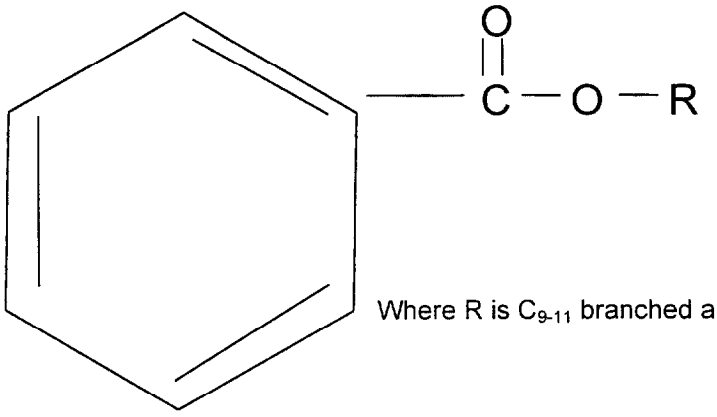

Where R is C_{9-11} branched alkane

Isodecyl Benzoate

CAS NO. 131298-44-7

USEPA HPV CHALLENGE PROGRAM SUBMISSION

November 21, 2001

Submitted by:

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TESTING PLAN

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EXECUTIVE OVERVIEW

Isodecyl Benzoate is a clear colorless liquid used primarily as a coalescent in water based paints and as a secondary plasticizer in PVC. It is typically incorporated at a level of 1 to 2% of the total paint formulation and at 5-8% as a secondary plasticizer in PVC.

The physical/chemical properties of isodecyl benzoate are adequately defined. It is a low volatility liquid at room temperature with a boiling point range of 321.5 to 342.5°C and a vapor pressure of 8.45×10^{-3} Pascals. Its partition coefficient is high (log K_{ow} = 4.61) and its solubility in water is low (<0.686×10^{-4} g/L). Isodecyl benzoate has a freezing point of -25^{0} C.

In the area of environmental fate and pathways, adequate studies were available to assess biodegradation but no information was available for photodegradation potential, water stability or transport and distribution in the environment. In 28-day studies, isodecyl benzoate attained a 20% biodegradation in one study (OECE 301D) and a 67%beidegradation in another study (OECD301C). Although little else is known about the environmental fate of isodecyl benzoate, its physical/chemical properties suggest that volatilization is not the dominant fate process controlling its distribution in the environment.

Relative to ecotoxicity, adequately conducted aquatic toxicity test studies consistent with OECD Guidelines have shown no effects on algal growth (96-hr EC50=50 g/L), the life-cycle of *Daphnia magna* (LOEC/MATC=39 g/L for 21 days) survival of the rainbow trout (96-hr LC50=>6.5 mg/L), or early life-stage development of the fathead minnow (LOEC/MATC=>47 g/L). However, the 48-hour EC50 for *Daphnia magna* in an acute study was 0.54 mg/L, suggesting a moderate-to-high degree of aquatic toxicity. This is particularly important in view of the questionable biodegradability of isodecyl benzoate.

Adequate acute toxicity studies consistent with OECD guideline have shown a low degree of toxicity by the oral route (Rat LD50=>5000 mg/kg), inhalation route (Rat 4-hr LC50=3.3mg/L), and dermal route (Rabbit LD50=>2000mg/kg). In addition, isodecyl benzoate produced slight-to-moderate eye and skin irritation in rabbits but was shown not to be an allergic skin sensitizer in guinea pigs. In a well-conducted 28-day repeated dose study (OECD 407), rats dosed by oral gavage showed behavioral changes and increased liver weights and liver histopathology at 1000mg/kg, but 150 mg/kg was the NOAEL and 15 mg/kg was the NOEL for the study. This data, too, suggests a low order of repeated-exposure toxicity. There was no gross or microscopic pathology on male or female reproductive organs in rats at <1000mg/kg. In a developmental toxicity study (OECD 415), slight suggestions of fetal retardation in association with maternal toxicity were reported at a dose of 1000 mg/kg. However, a 300 mg/kg dose was considered to be the NOAEL for this study for both maternal and fetal effects. Isodecyl benzoate does not appear to pose a unique hazard to the developing fetus.

Relative to genetic toxicity potential isodecyl benzoate was not mutagenic in several *in vitro* and *in vivo* studies (consistent with OECD Guidelines) measuring both point mutation and chromosome aberration endpoints of concern for HPV purposes. Isodecyl benzoate produced negative results in two bacterial reverse mutations assays, in a human lymphocyte assay, and in a mouse micronucleus study.

With regard to the EPA/HPV Program, Velsicol Corporation has determined that no additional testing is needed in the areas of PHYSICAL/CHEMICAL PROPERTIES, ECOTOXICITY, and MAMMALIAN TOXICITY. However, in the area of ENVIRONMENTAL FATE AND PATHWAYS, only biodegradation studies consistent with OECD Guidelines satisfy the HPV requirements. Therefore, Velsicol Corporation proposes to conduct studies to evaluate photodegradation potential, stability in water, and transport and distribution in the environment. The preceding studies (tests or modeling) to meet HPV requirements will be consistent with OECD Guidelines and will be conducted in 2002.

Testing Plan and Rationale

TESTING PLAN IN TABULAR FORMAT

Isodecyl Benzoate CAS No. 131298-44-7 or 120657-54-7	Information Available?	OECD Study?	GLP Study?	Other Study?	Estimation Method?	Acceptable?	Testing Recommended?	Comments
HPV Endpoint								
Physical/Chemical Properties								
Freezing Point	Y	N	Y	N	N	Y	N	
Boiling Point	Y	N	Y	N	N	Y	N	
Vapor Pressure	Y	N	Y	N	N	Y	N	
Partition Coefficient	Y	Y	Y	N	N	Y	N	
Water Solubility	Y	N	Y	N	N	Y	N	
Environmental Fate								
Photodegradation	N						Y	
Water Stability	N						Y	
Transport	N						Y	
Biodegradation	Y	Y	Y	N	N	Y	N	
Ecotoxicity								
96-Hour Fish	Y	Y	Y	N	N	Y	N	
48-Hour Invertebrate	Y	Y	Y	N	N	Y	N	
72-Hour Algae	Y	Y	Y	N	N	Y	N	
MammalianToxicity								
Acute Toxicity	Y	Y	Y	N	N	Y	N	
Repeated Dose	Y	Y	Y	N	N	Y	N	
Genotoxicity (Point Mutation)	Y	Y	Y	N	N	Y	N	
Genotoxicity (Chromosome Aberration)	Y	Y	Y	N	N	Y	N	
Reproductive/Developmental Toxicity Endpoint	Y	Y	Y	N	N	Y	N	

INTRODUCTION

Isodecyl benzoate (CAS No.131298-44-7 or 120657-54-7) is an alcohol used paints and PVC. In water based paints, it is used as a coalescing agent and incorporated at a level of 1 to 2% of the total paint formulation. It is also used as a secondary plasticizer in PVC where it may comprise 5-8% of the PVC product.

Since isodecyl benzoate is a high-boiling liquid of very low volatility (vapor pressure of 8.45 $x10^{-3}$ Pascals), little or no vapor exposure occurs in the industrial setting. Although this material is not absorbed through the skin in toxicologically significant amounts, dermal exposure is also kept to a minimum in occupational settings since isodecyl benzoate can produce mild-to-moderate skin and eye irritation. Since there are few sites of manufacture, the number of potentially exposed workers is also small and no occupational exposure limit for workplace air has been proposed for this material.

Various studies consistent with OECD Guidelines have been conducted on isodecyl benzoate. These studies are briefly summarized in this rationale document describing whether or not they meet the requirements of the EPA/HPV Program. Robust summaries, using a SIDS format, have been prepared for key studies (and some supporting studies) and are included as an appendix to this document.

PHYSICAL/CHEMICAL DATA

Physical/chemical data for isodecyl benzoate are available from studies using protocols consistent with OECD guidelines:

- Freezing Point — -25^{0}C (1)
- Boiling Point/Range — 321.5 to 342.5^{0} C (2)
- Vapor Pressure — $8.45x10^{-3}$ Pascals (3)
- Partition Coefficient — Log K_{ow}=4.61 (4)
- Water Solubility — <$0.686x10^{-4}$g/L (5)

These properties indicate that isodecyl benzoate is a liquid with a very low vapor pressure and a very low water solubility. Its relative density (D20/4) is 0.95155 (6). In addition, isodecyl benzoate has a flashpoint of 110°C (7) and an autoignition temperature of 374^{0}C (8). It is not flammable in contact with water (9), not pyrophoric (10) and not explosive (11).

Recommendation: No additional studies are recommended. The available data fulfill the HPV required endpoints.

ENVIRONMENTAL FATE AND PATHWAYS

No information was available on isodecyl benzoate relative to photodegradation, stability in water (hydrolysis), or transport and distribution in the environment. Two different biodegradation studies following OECD guidelines were conducted. In a ready biodegradability study (OECD 301D) (12), isodecyl benzoate attainted only 20% biodegradation after 28 days and therefore cannot be termed as biodegradable in this closed bottle test. However, in another biodegradation study using a modified MITI test (OECD 301C) (13), isodecyl benzoate was readily biodegradable under the test conditions; its percentage biodegradability by BOD was 67% after

28 days. In one additional study measuring respiration inhibition (OECD 209) (14), the 3-hour EC50 was greater than 100mg/L, the highest test concentration that could be prepared due to limited solubility in water.

Recommendation: Since only the preceding biodegradation studies are adequate to meet HPV requirements, isodecyl benzoate will be tested for photodegradation potential (using AOPWIN v1.90 SAR model), stability in water (OECD 111 or Estimation Model), and transport and distribution (Fugacity Modeling using Level III Mackay-type methods).

ECOTOXICITY

Isodecyl benzoate was found to have a 96-hour LC50 of >6.5mg/L in rainbow trout (OECD 203) (15). Although a nominal concentration of 100mg/L was used for the study, the measured test concentration (6.5mg/L) was the highest that could be attained based on the very low water solubility of the test substance. No mortality, adverse clinical signs or abnormal behavior were noted during the 96-hour exposure period. Therefore, the NOEC and the No-Mortality-Concentration was reported as 6.5mg/L.

Aquatic invertebrate toxicity was examined in a study (OECD 202) of *Daphnia magna* (16). Isodecyl benzoate was tested at measured concentrations of 0.089, 0.11, 0.28, 0.46 and 0.70 mg/L. The 48-hour EC50 was calculated to be 0.54 mg/L. Deaths occurred at the two highest exposure levels and the No-Mortality-Concentration was 0.28mg/L. The NOEC for the study was 0.089 mg/L.

Toxicity to aquatic plants was evaluated in freshwater algae (*Selenastrum capricornutum*) in a static test consistent with OECD guidelines (17). Isodecyl benzoate was tested at measured concentrations of 6.1, 16 and 50 g/L. There were no statistically significant effects on mean cell

density or cell growth in any test group. Therefore, the 96-hour EC10, EC50 and EC90 for both cell density and cell growth were reported at >50 g/L.

Two chronic toxicity tests were also conducted in aquatic organisms. In an early life-stage toxicity test with fathead minnows (18), isodecyl benzoate was tested at nominal concentrations of 0.81, 2.7, 9.0, 30 and 100 g/L for 33 days (5-day embryo hatching period and a 28-day post-hatch juvenile growth period). Analysis of the 3 lowest levels indicated concentrations below the limit of quantification (6 g/L) and the mean measured values for the two highest concentrations were 13 g/L and 47 g/L. No adverse effects were seen relative to hatching success survival or growth at any of the concentrations tested. In a life-cycle toxicity test with *Daphnia magna* (19), daphids were exposed to isodecyl benzoate at nominal concentrations of 0.81, 2.7, 9, 30 or 100 ug/L in a continuous-flow diluter system for a 21-day study period. Chemical analysis indicated that the mean measured concentrations for the three lowest concentrations were less than the limit of quantification (6 g/L); measured concentrations for the two highest concentrations were 10 and 39 g/L). There were no adverse effects on survival, reproduction or growth at any dose level. Therefore, the LOEC and the maximum acceptable toxicant concentration (MATC) were both >39 g/L.

In summary, ecotoxicity showed no effects on algal growth (50 g/L), life cycle of *Daphnia magna* (39 g/L), survival of rainbow trout (6.5 mg/L), or early life-stage development of the fathead minnow (47 g/L). However, the EC50 (48 hours) in a toxicity test with *Daphnia magna* was 0.54 mg/L, indicating a high degree of toxicity to this aquatic organism, particularly in view of the fact that the material may not be readily biodegradable.

Recommendation: The ecotoxicity studies assessing acute toxicity potential are adequate to meet HPV requirements. No additional testing is recommended.

MAMMALIAN TOXICITY

A. Acute Toxicity

The acute oral toxicity of isodecyl benzoate was determined in a study (OECD 401) on 5 male and 5 female Sprague-Dawley rats using the neat material given by gavage at a concentration of 5000mg/kg body weight. No mortality occurred during the 14-day post-dosing period although two rats of each sex had diarrhea 4 hours after dosing and all rats showed a yellow-stained genital area. The LD50 was reported a >5000mg/kg (20).

The acute inhalation toxicity potential of Isodecyl Benzoate was determined in a study (OECD 403) on Sprague-Dawley rats (5/sex/concentration) at respirable aerosol concentrations of 1, 3 and 5 mg/L, respectively, administered over a 4-hour exposure period. For the combined sexes, the 4-hour LC50 was calculated to be 3.3 mg/L. Adverse clinical signs during exposure at all test levels included respiratory difficulty (dyspnea, polypnea), squinting, tremors and hunched appearance during exposure. Between 50 minutes and 5 days post-exposure, mortality occurred at the mid-and high-test levels. During the second post-exposure weels, all survivors appeared normal except for occasional sores and alopecia (21).

Isodecyl benzoate also produced no mortality in a dermal toxicity study (consistent with OECD guidelines) where 5 male and 5 female rabbits were dosed at 2000mg/kg body weight as the neat material. The dermally-applied dose remained in contact with the skin for 24 hours and animals were observed for 14 days thereafter. No mortality or adverse clinical signs occurred during the study. Dermal irritation did occur at the site of application and consisted of edema, erythema and desquation, slight atonia, coriaceousness and fissuring. The dermal LD50 in rabbits was reported as >2000 mg/kg (22).

Isodecyl benzoate also produced slight-to-moderate erythema and edema in rabbits in a dermal irritation study, but it is not classifiable as a skin irritant according to OECD guidelines (23). It also produced slight-to-moderate conjunctival irritation in rabbits in an eye irritation study (24) but again, it is not classifiable as an eye irritant according to OECD guidelines. Finally, in both a modified Buehler test (25) and a maximization test (26) in guinea pigs, isodecyl benzoate was not an allergic skin sensitizer.

Recommendation: All acute toxicity studies were consistent with OECD guidelines and meet HPV requirements. No additional acute toxicity testing is recommended.

B. Repeated Dose Toxicity

In a repeated-dose oral gavage study (consistent with OECD 407), Sprague-Dawley rats (8/sex/dose) were dosed with isodecyl benzoate for 29 consecutive days at concentrations of 0, 15, 150 and 1000mg/kg body weight in corn oil. In addition to all required measurements, a functional observational battery (FOB) was performed on all animals before testing and during Week 2 and Week 4 of dosing. At 1000mg/kg, behavioral changes, suggestive of an effect on the nervous system, were seen. High-dose animals also had increased liver and kidney weights, centrilobular hepatocyte enlargement, and eosinophilic intracytoplasmic droplets in the proximal convoluted tubules (males rats only). The latter finding was the only treatment related effect seen at the 150 mg/kg dose in male rats. This type of kidney finding is specific to male rats and is not considered predictive for a similar effect in man. No adverse effect was seen at 15 mg/kg. Therefore, for this study, 150 mg/kg was considered to be the NOAEL and 15mg/kg was the NOEL (27).

Recommendation: This repeated dose study meets HPV requirements and no additional testing is recommended.

C. Genetic Toxicity

The SIDS/HPV requirements for genetic toxicity testing are for two endpoints: one sensitive to point mutation and one sensitive to chromosomal aberrations. Isodecyl benzoate studies subsequently described (in vitro and in vivo) fulfill those requirements.

A *Salmonella typhimurium* reverse mutation assay (OECD 471) was conducted in 5 strains of bacteria in triplicate, with and without metabolic activation, at concentrations of isodecyl benzoate ranging from 50 to 5000 g/plate. No mutagenic response was recorded in this study (28). This study is supported by a negative finding in another limited *Salmonella* assay conducted earlier at isodecyl concentrations up to 10,000 g/plate (29). In an *in vitro* study sensitive to chromosome aberrations (OECD 473), cultured human lymphocytes were exposed to isodecyl benzoate, with and without metabolic activation, at concentrations ranging from 19.5 to 5000 g/ml. No evidence of clastogenic activity was seen in this *in vitro* cytogenetic test system (30).

In an *in vivo* mouse micronucleus study (OECD 474), male and female mice (Swiss SPF CD-1 Outbred) were given a single intraperitoneal dose of 1280 mg/kg isodecyl benzoate (maximum tolerated dose). Bone marrow smears from 5 male and 5 female mice in the negative control (aqueous 1% methylcellulose) and the test substance groups were obtained at 24, 48 and 72 hours after dosing. At all sampling times, mice treated with isodecyl benzoate showed no significant increase in the frequency of micronucleated polychromatic erythrocytes. Thus, there was no evidence of chromosome damage in this *in vivo* test (31).

Recommendation: The preceding *in vitro* and *in vivo* genotoxicity tests are adequate and meet HPV requirements. No additional genotoxicity studies are recommended.

D. Reproductive Toxicity

In accordance with OECD Guideline 407, a 28-day repeated-dose, oral gavage study (27) (Described earlier under REPEATED DOSE TOXICITY) on isodecyl benzoate also included a thorough gross and microscopic examination of male and/or female reproductive tissues: ovaries, epididymides, prostate, seminal vesicles and testes. For the microscopic examination, 5 rats/sex from the high dose (1000mg/kg) and controls were examined - ovaries and testes with epididymides. There was no adverse effect on reproductive organ weights and the histopathology exam on ovaries and testes was unremarkable. An adequate repeated-dose study (without a mating trial), such as this 28-day study, in conjunction with an adequate developmental toxicity study (see DEVELOPMENTAL TOXICITY Section of this document), should be considered acceptable to fulfill the reproductive/developmental toxicity endpoint for both the OECD/SIDS Program and the HPV Program.

Recommendation: No additional studies are recommended.

E. Developmental Toxicity

In an oral gavage study (OECD 414 (32), 25 pregnant Sprague-Dawley Crl:CD®BR rats/dose were given isodecyl benzoate at doses of 0 (corn oil), 30, 300 and 1000 mg/kg body weight on days 6 through 15 of gestation. At the highest dose (1000mg/kg), only minimal adverse effects were seen –a transient decrease in body weight gain in maternal rats, and a decrease in mean fetal weight and a slight reduction in the incidence of cervical centrum mo. 1 ossified; the later two findings are suggestive of developmental retardation in the fetuses. No other treatment-related malformations or developmental variations were observed at any dose level. A dose of 300mg/kg was considered to be the NOAEL for both maternal and developmental toxicity. A dose of 30mg/kg was the NOEL for this study. Since only minimal developmental effects were seen in

the fetuses at 1000mg/kg, a dose that also produced maternal toxicity, isodecyl benzoate was considered to pose no unique hazard to the developing fetus.

Recommendation: In conjunction with the negative findings on reproductive organs in the 28-day repeated dose study at ≤1000mg/kg in both male and female rats, this developmental toxicity study fulfills both OECD/SIDS and HPV requirements. No additional testing is recommended.

CONCLUSIONS

Overall, isodecyl benzoate does not appear to represent an unacceptable risk to human health. Under the EPA/HPV Challenge Program, isodecyl benzoate was evaluated and data gaps were identified for ENVIRONMENTAL FATE AND PATHWAYS only. In particular, photodegradation potential, water stability, and transport and distribution in the environment will be determined in studies (tests or estimation modeling) that reference OECD Guidelines. With regard to other parameters specified in the EPA/HPV Challenge Program, the available data fill all of the requirements for physical/chemical properties, ecotoxicity, and mammalian toxicity (acute and repeated dose toxicity, genotoxicity, and reproductive/developmental toxicity). Since no studies using animals have been recommended, we feel that animal welfare concerns have been properly addressed. Appropriate studies to meet HPV requirements will be conducted in the first quarter of 2002 and take about 6 months to complete.

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